

## ***Sclerotinia* resistance in sunflower: I. Genotypic variations of hybrids in three environments of Argentina**

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### **Summary**

*Sclerotinia sclerotiorum* infections on sunflower capitula produce white rot, one of the most dangerous diseases sunflower bears in all humid areas. Therefore, disease quantification on developed genetic materials is a very important action in sunflower breeding. Given the horizontal type of resistance these evaluations should be made in different environments. Several hybrids obtained after crossing seven female  $\times$  seven male sunflower inbred lines in a factorial mating design were evaluated for resistance to white rot in two locations in the southeast of the province of Buenos Aires, Argentina. Disease incidence and the symptom appearance index indicated significant genotypic effects and genotype  $\times$  environment ( $G \times E$ ) interactions. Genotypic effects were a greater source of variance than the  $G \times E$  interaction effects. The  $G \times E$  interactions only represented changes in magnitude. These results were used to identify the best genotypes for both resistance factors. Four hybrids showed low disease incidence values under both artificial and natural infections, of which only two had high values of the symptom appearance index at both locations. These hybrids are considered to have adequate resistance attributes for the ascospore penetration and the mycelium extension in the capitulum tissue and could therefore be sown in the province of Buenos Aires without increasing risks of *S. sclerotiorum* attacks.

### **Introduction**

Sunflower (*Helianthus annuus* L.) is the most important annual species grown worldwide specifically for its edible oil. In Argentina, sunflower seed production was 3.7 million tons in 2002–2003 (USDA, 2004), which makes it the leading producer. Argentinean farmers grow sunflower in an area involving a wide range of agro-climatic situations, although over 60% of the national seed production is located mainly in the province of Buenos Aires (Devoto & Guibert, 2000). This normally has temperate and humid conditions during the cropping season.

*Sclerotinia sclerotiorum* (Lib.) de Bary is a common and widespread pathogen in sunflower crops

grown in humid and temperate areas in the world. It attacks different plant organs such as roots, basal and mid-stem, buds, leaves and inflorescences (Castaño et al., 1989). The infections on capitula produce white rot a disease considered as the main factor that determines the success of the crop either because of a partial seed yield reduction (Sadras et al., 2000) or a total crop destruction (Gulya et al., 1997). White rot can also reduce sale allowances, as the presence of sclerotia in seed lots increases seed acidity, thereby reducing seed quality (Agüero et al., 1997). All of this makes white rot the most studied disease in sunflower (Vear, 2004).

Most sunflower farmers grow single-cross hybrids. In Argentina there are several private and public

sunflower seed companies interested in developing this type of cultivars. In breeding plans for sunflower hybrids one of the principal characters to be selected is white rot resistance. Given the quantitative nature of white rot resistance in sunflower (Castaño et al., 2001a,b), this is a difficult and onerous task because of a need to conduct trials across environments to accurately measure the genotypic value of genetic materials.

Genotypic values of hybrids can be affected by environmental variability. In sunflower, there are several reports showing changes of genotypic values due to environmental effects on some agronomic and morpho-phenological attributes (Alza & Fernández-Martínez, 1997; Razi & Assad, 1999; de la Vega et al., 2001; Foucteau et al., 2001; Lúquez et al., 2002) as well as on disease responses to *Verticillium* wilt (Escande et al., 2000) and to *Phomopsis* stem canker (Vigué et al., 2000). All these authors suggested that the alteration of genotypic values reduces the correlation between the genotype and phenotype values and therefore complicates the breeding and testing of improved genetic materials.

Studies on the evaluation of genotypes for white rot resistance in different environments are scarce with only a few reports and that only for European conditions. In France, Tourvieille et al. (1996) and Serre et al. (2004) evaluated one hundred of F3:4 lines in six locations and two inbred lines over 13 years. In both studies mid-correlation coefficients were detected between genotypes across environments. In Germany, Hahn (2002) showed a significant genotype  $\times$  year interaction variance and a moderate correlation value between years for 45 inbred lines of diverse origin.

No studies have been reported in Argentina on the performance of sunflower hybrids across environments against the *S. sclerotiorum* infections on capitula. The results of such a work would help breeders to improve the description of genotypic responses to different factors of white rot resistance. In addition, the need for dividing the large sunflower growing region of the province of Buenos Aires, Argentina, with the purpose of exploiting specific adaptations to *S. sclerotiorum* resistance could thus be considered.

The objectives of this investigation were then to evaluate the performance of a set of sunflower hybrids grown in two experimental sites of the province of Buenos Aires, Argentina, under *S. sclerotiorum* infections, and to identify genotypes showing an adequate level of resistance on capitula.

## Materials and methods

### *Plant material*

Forty-nine sunflower F1 hybrids were developed by crossing seven cytoplasmic male-sterile female inbred lines (i.e., A-lines) to seven fertility-restorer male lines (or R-lines) in a factorial design. Six inbred A-lines (ADK1, ADK2, ADK3, ADK4, ADK5, and ADK6) and 6 R-lines (R122, R152, R161, R167, R199, and R200) were provided by the sunflower breeding program of Monsanto Argentina. All these lines were originated from the introgression of the Argentinean gene pool into the old Russian materials improved in the USA. The remaining two lines, SD (A-line) and PAC1 (R-line), were developed at INRA, France, from Russian, Romanian and North-American sunflower populations. Inbred lines from Monsanto are a random sample of the company's available genetic variability used in the development of sunflower hybrids and they are neither inter-related nor related to the French lines (J. Ré, personal communication). The non-commercial hybrid SCKR, moderately resistant to *S. sclerotiorum* infections on capitula (Álvarez et al., 1999), was used as a check in infection quantification.

### *Experimental design*

Field experiments were conducted in Camet and Balcarce, two sites located in the southeast of the Buenos Aires Province. They are 75 km apart and Camet has a more marked marine climate because of its proximity to the Atlantic Ocean. Hybrids were grown in two experiments (E1 and E2) at Camet and one (E3) at Balcarce. Materials planted in E2 had a high probability of natural infection by *S. sclerotiorum* as the soil site had been infested with the pathogen's sclerotia.

Experiments were in a randomized complete block design, with three replications for E1 and E3 and two for E2 and had different sowing dates. Each plot had at least 15 plants. To obtain staggered flowering dates of the control, the SCKR check was sown in E1 and E3 on three dates at intervals of seven days each without randomization.

### *Inoculum and inoculation method*

Each experiment site used its own inoculum. Sclerotia were harvested in Camet and Balcarce from sunflower plants naturally attacked by *S. sclerotiorum* the year before experiments were conducted. Ascospores

were obtained following the Castaño and Rodríguez's (1997) procedure. Artificial infections were made on all sunflower capitula of E1 and E3, when their three external rows of hermaphrodite disk flowers were in pistillate stage (R5.3) (Schneider & Miller, 1981) or in their homologous stage (F3.2) (Cetiom, 1992). The floral surface of each capitulum was sprayed with 5 ml of an aqueous suspension containing approximately 5000 ascospores ml<sup>-1</sup>. Infections were carried out twice a week, each plant being infected once at the correct stage. At each infection date, 20 plants of the SCKR check were similarly infected. In E3 the infected capitula were immediately covered with Kraft paper bags and a commercial sprinkler overhead irrigation system was used at a rate of 5 mm twice per week until crop maturity. In E1, the capitula were kept uncovered and sprayed with water two or three times a day by an overhead micro-sprinkler system until maturity. In E2, capitula were also uncovered but were not sprayed.

Starting at three weeks after inoculation each capitulum in E1 and E3 was observed twice a week, and the dates of first white rot symptoms were recorded. Two variables were evaluated according to Castaño et al. (1993): the disease incidence (DI) calculated at maturity for each hybrid as: [number of capitula showing symptoms after inoculation/number of inoculated capitula] × 100. The second was the symptom appearance index (SAI) calculated as the plot mean value of the incubation index (e.g., incubation period for hybrids/incubation period of the SCKR check inoculated at the same date) of each inoculated capitulum. SAI measures the delay (in days) from inoculation to symptom appearance relative to controls; therefore the SAI increases with increasing resistance to *S. sclerotiorum*. In E2, where the dates of natural infections were not known hence incubation periods could not be determined, only the proportions of white rotted capitula were scored, on a single date, and at crop maturity.

#### Statistical analyses

Two hybrids (ADK3 × R200, ADK5 × R199) were excluded from analysis because of seed germination problems. Data from E2 were modified following the square root of the relative number of capitula showing symptoms (%) + 0.5 transformation ( $\sqrt{\% + 0.5}$ ).

Analyses of variance were made individually for each experiment. The variance error equality in E1 and E3 was checked by the Bartlett's test for homogeneity, followed by a nested analysis of variance combined

over locations. A two-factor mixed model (fixed hybrid genotypes and random environments) was used following Romagosa and Fox (1993). *F*-test was made to detect differences between sources of variation. The test of the least significance difference (LSD) was used to determine dissimilarities among hybrid responses, and three groups of different levels of resistance were constituted. Two of them contained hybrids in which estimated disease values were statistically similar to either the maximum or the minimum disease values obtained in the experiments; hybrids placed in the third group differed from those ones having the maximum and minimum disease values. A series of orthogonal contrasts were computed to make planned comparisons between the disease mean values shown by the same hybrid in two environments. The Spearman's rank correlation test was used to evaluate the degree of association between the ranks of hybrid responses after pathogen infections in two locations. Statistical parameters were estimated according to Sokal and Rohlf (1981).

## Results

### Artificial infections

#### Disease incidence

The range of disease incidence (DI) in sunflower hybrids after *S. sclerotiorum* infections at Camet (E1) and Balcarce (E3) was 93% (Table 1). The error variances within experiments (locations) were homogeneous as was indicated by Bartlett's test.

The combined analysis of variance did not detect environmental effects. All dissimilarities in the management practices before and after *S. sclerotiorum* infections at each site (i.e., sowing dates, irrigation methods, pathogen inoculums, paper bag utilization) as well as some of the meteorological variables (e.g., temperature, rainfall, radiation, relative humidity) that might have exist between the locations had no significant effect on the environmental means (i.e., Camet = 67% and Balcarce = 58%).

Highly significant effects of hybrids and genotype × environment (GE) interaction were detected. The variability of the GE interactions was 54% lower than that corresponding to hybrid effects. Differences in the relative performances of individual hybrids for DI in E1 and E3 suggest that genotypes and environments were not independent factors; therefore the hybrid performance should be evaluated and analyzed in different environments.

Table 1. Disease incidence (DI) (%) in sunflower hybrids after *S. sclerotiorum* artificial infection on capitula in Camet (E1), in brackets, and in Balcarce (E3) and orthogonal differences of each hybrid in these two experimental sites

Female lines	ADK1	ADK2	ADK3	ADK4	ADK5	ADK6	SD
Males lines							
R122	[98] 72	[97] 89	[92] 64*	[96] 72	[100] 80	[69] 58	[83] 55*
R152	[100] 82	[37] 87**	[40] 41	[44] 84**	[95] 80	[55] 80	[7] 61**
R161	[83] 59	[44] 60	[33] 41	[69] 47	[96] 57**	[52] 28	[43] 71*
R167	[96] 62*	[70] 73	[62] 30*	[74] 88	[100] 65*	[52] 40	[38] 84**
R199	[83] 36**	[40] 29	[26] 16	[62] 80	—	[28] 26	[22] 21
R200	[100] 50**	[88] 48**	—	[91] 72	[100] 58**	[70] 58	[77] 33**
PAC1	[85] 73	[46] 60	[72] 46	[33] 37	[96] 84	[79] 51*	[25] 43

Note. General mean = 62%; CV = 26%; F hybrids = 9.13\*\*; F hybrid-location interaction = 4.23\*\*;  
LSD<sub>0.05</sub> = 23% (E1) and 28% (E3).

\* $p < 0.05$ , \*\* $p < 0.01$ , respectively.

In Camet (E1), the LSD value permitted classification of genotypes into three groups. Group 1 (G1) had 20 hybrids of which five had the maximum DI values in E1; given the high proportion of diseased capitula this is the group with the lowest resistance in this study. Four hybrids had the DI values similar to that of SD×R152, which has the minimum value (7%); these five hybrids constitute Group 3 (G3), the group with the highest resistance. The remaining 22 hybrids were included into Group 2 (G2), with the level of resistance intermediate between G1 and G3.

Similar three groups have also formed at Balcarce (E3). Nineteen hybrids had DI values similar to the hybrid with the maximum value, and all 20 were in the susceptible group G1. Twelve hybrids were similar to hybrid ADK3×R199 and they were assigned to the resistant group G3. The remaining 14 hybrids were in the intermediate resistant G2 group.

The hybrid SD×PAC1 showed DI values of 25 and 43% at E1 and E3, respectively, indicating that its performance was not different from the hybrids with the minimum DI value. Therefore, SD×PAC1 was assigned to the G3 group of low proportion of diseased capitula in each experiment. This hybrid is well-known in France as a white rot resistant check (Vear and Tourvieille, 1988; Castaño, 1992). Our results indicate that SD×PAC1 can be also used as a resistant check in Argentina.

The Spearman's coefficient of correlation value was highly significant ( $r_s = 0.40$ ,  $p < 0.01$ , 47 d.f.), and shows an agreement in the genotypic ranks in E1 and E3. This result indicates the presence of a proportionate genotypic response or non-crossed interactions of hybrids between locations.

The orthogonal contrasts analysis has shown that 17 hybrids had significant differences in their DI values (Table 1) in the two locations. This maybe responsible for the detected GE interaction effect in the two locations. Of these 17 hybrids, 12 from Camet showed higher DI values compared to Balcarce and five from Balcarce showed higher DI values compared to Camet.

#### Symptom appearance index

The SAI values were calculated from more than ten diseased capitula per line (Castaño et al., 1993) except for SD×R152 and SD×PAC1, which had only three and eight diseased capitula, respectively, in Camet, and SD×R199 and ADK3×R199, with nine and seven diseased capitula, respectively, in Balcarce. The SAI values ranged from the minimum of 0.58 for ADK5×R122 and ADK5×R161 (the most susceptible hybrids) to the maximum of 1.68 for ADK1×R199 (the most resistant hybrid) (Table 2). According to the Barlett's test error variances in both experiments were homogeneous.

The combined analysis of variance showed no significant differences between location means (i.e., Camet = 0.96 and Balcarce = 1.12) but highly significant differences were detected for hybrids and GE interaction effects. The variability of the GE interaction effects was however 67% lower than that corresponding to hybrids. This suggests that hybrids had to be characterized for SAI in each environment, as the relative performance of hybrids was different in E1 and E3. In Camet, based on the LSD test value, three groups were formed, similarly to DI. Two hybrids had SAI values similar to the lowest one, and they formed G1; this is the group of the lowest resistance in this study, and

Table 2. Symptom appearance index (SAI) in sunflower hybrids after *S. sclerotiorum* artificial infection on capitula in Camet (E1), in brackets, and in Balcarce (E3) and orthogonal differences of each hybrid in these two experimental sites

Female lines	ADK1	ADK2	ADK3	ADK4	ADK5	ADK6	SD
Male lines							
R122	[0.66] 0.82	[0.75] 0.65	[0.77] 0.96	[0.80] 1.10*	[0.58] 0.70	[0.91] 0.91	[0.89] 1.46**
R152	[0.84] 0.99	[0.88] 1.14	[1.11] 1.38	[0.98] 1.09	[0.77] 0.83	[1.00] 0.94	[1.45] 1.41
R161	[1.01] 1.26	[0.99] 1.05	[0.93] 1.10	[1.06] 1.19	[0.77] 0.58	[1.05] 1.32	[1.10] 1.06
R167	[0.82] 1.13*	[0.99] 1.05	[1.01] 1.32*	[1.14] 1.38	[0.73] 0.83	[1.01] 1.31	[1.10] 1.33
R199	[0.90] 1.68**	[1.02] 1.19	[1.24] 1.08	[0.98] 1.48**	–	[1.12] 1.56**	[0.96] 1.40**
R200	[0.86] 0.92	[0.90] 1.09	–	[0.98] 1.16	[0.79] 0.97	[0.98] 0.86	[0.99] 1.16
PAC1	[0.90] 1.02	[1.04] 1.01	[1.13] 1.05	[1.13] 1.32	[1.03] 0.93	[1.06] 1.08	[1.00] 1.32*

Note. General mean = 1.03; CV = 17%; F hybrids = 5.87\*\*; F hybrid-location interaction = 1.83\*\*; LSD<sub>0.05</sub> = 0.16 (E1) and 0.38 (E3).

\*  $p < 0.05$ ; \*\*  $p < 0.01$ .

these hybrids manifested the first symptoms a few days after the infection date. The G3 had only one hybrid (SD×R152), which showed the latest appearance of white rot symptoms; whereas the remaining 43 hybrids were in the intermediate G2 group. In Balcarce (E3), the G1 group had ten hybrids, which did not differ from the hybrid with minimum value. In the G3 group, 13 hybrids did not differ from ADK1×R199, which showed the maximum value. Finally, the G2 group comprised 22 hybrids.

For SAI, the hybrid SD×PAC1 was placed in the G2 and G3 groups in Camet and in Balcarce, respectively. This result, particularly that of Balcarce, is close to that one estimated by Vear and Tourvieille (1988) in France; this suggests again a similar behavior of the hybrid in different environmental conditions.

The Spearman's correlation value of  $r_s = 0.55$  (d.f.47), highly significant ( $p < 0.001$ ), indicates that non-crossed interactions were identified again because a comparable ranking of hybrids was detected in both locations for SAI.

The orthogonal contrasts analysis showed that nine hybrids had significantly higher SAI values in Balcarce than in Camet (Table 2) because the relative number of days without white rots increased in E3. This quantitative change in magnitude of the level of resistance in these hybrids could be contributed to the non-crossed interaction effects across locations for SAI.

#### Natural infections

Table 3 shows mean values of the proportion of white rotted capitula on sunflower hybrids under natural infection in E2. The range of the proportion of diseased

Table 3. Proportion of infected capitula in sunflower hybrids after *S. sclerotiorum* natural infection in Camet (E2)

Female lines	ADK1	ADK2	ADK3	ADK4	ADK5	ADK6	SD
Male lines							
R122	42	20	13	13	69	13	2
R152	17	7	2	11	43	16	2
R161	41	27	4	13	35	0	5
R167	4	15	6	35	33	27	0
R199	15	14	2	0	–	7	15
R200	11	5	–	16	19	2	7
PAC1	5	9	2	10	18	4	18

Note. General mean = 15%; CV = [46]; F hybrids = [2.81]\*\*; LSD<sub>0.05</sub> = [3.12]; [...] = results using transformed data  $\sqrt{\% + 0.5}$ . \*\*  $p < 0.01$ .

capitula was 69%. The estimated coefficient of variability suggests a relatively high heterogeneity of the data. The heterogeneous distribution of the pathogen in the soil, the variability of the ascospore number arriving on the capitula and the lack of control in the environmental humidity for the appearance of disease symptoms, might be some of the variables determining the high variability of the recorded data. In spite of this, the moderate precision of data agrees with those ones shown by Vear and Tourvieille (1988) and Achbani et al. (1994), when sunflower hybrids were naturally infected on capitula and terminal buds, respectively. According to these results, it seems that this level of non-controlled variability in this type of experiments is quite common.

Analysis of variance detected statistically significant differences among genotypic responses. Seven hybrids showed a relative number of diseased capitula

similar to the hybrid with the maximum value in E2; they formed the susceptible G1 group. Seventeen hybrids had values similar to those ones showing the minimum values (SD×R167, ADK6×R161, and ADK4×R199) and they are in the resistant G3 group. The remaining 19 hybrids are in the intermediate resistant G2 group.

The hybrid SD×PAC1 was placed in G2 group. Its relative number of diseased capitula recorded in E2 was similar to the ones obtained in France under natural infection by Vear and Tourvieille (1987, 1988). This result suggests that the good performance of this hybrid is repeatable in different environments, which makes it a very interesting hybrid to be included as a check in similar conditions of infection.

#### *Relationship between genotype responses under artificial and natural infections*

The DI mean performances of hybrids in E1 and E3 (Table 1) and those ones estimated in E2 (Table 3) were used to evaluate the association between hybrid responses to artificial and natural infections.

The Spearman's coefficient of correlation was highly significant ( $r_s = 0.46$ ,  $p < 0.001$ ), thus indicating that the ranking of hybrids obtained in both types of infections was quite associated. This result agrees with the discussion made by Tourvieille and Vear (1984) and Vear and Tourvieille (1988) in that the artificial infection test could be a good predictor of genotype performance under natural infection.

## **Discussion**

In this study the relative performance of a group of sunflower hybrids evaluated for resistance to *S. sclerotiorum* infections was assessed at three environments. This is the first time that this type of genetic material is evaluated as well as reported from Argentina.

Hybrid performance was evaluated by considering the two factors involved in white rot resistance for sunflowers (Castaño et al., 1993). The first factor was the ascospore penetration, quantified as a measure of percentage of infected plants (DI). The second factor was the rate of mycelial extension in adult tissues and was quantified by the relative incubation period (SAI).

A sunflower crop more resistant to *S. sclerotiorum* infections on capitula shows a lower number of diseased plants than a highly susceptible one (Castaño et al., 2001a). But a long incubation period is also

required for resistant hybrids. If infected plants showed their first white rot symptoms late, closer to the physiologic maturity stage, there would be a shorter period of time left in which diseased capitula could be destroyed before harvest (Russi et al., 2004). In addition, the period of sclerotia production would be also reduced. In this sense, it has to be highlighted that the appearance of first symptoms was the only resistance trait which showed genetic progress when a group of Argentinean hybrids released from 1973 to 2000 in our country was analyzed for white rot resistance (Castaño et al., 2003).

The scoring of both resistance factors simultaneously resulted in a better description of the hybrid responses to *Sclerotinia* attack in capitula and allowed the selection of genotypes possessing resistance genes for these two traits.

In our study, environments were considered as a random effect because the breeder has no control over the climatic conditions that will occur at any location in a year. In this sense, in spite of having different locations, the environmental means of the experiments E1 and E3 were statistically non-significant for DI and SAI traits. However, the variability shown by these two resistance factors was different depending on the location considered. In Camet, the environmental conditions magnified the DI responses in comparison with those ones detected at Balcarce, as the maximum (100%) and the minimum (7%) incidence values were reached at E1. On the other hand, both locations showed the same minimum value of SAI (0.58) but the maximum (1.68) was found in Balcarce. These results agree with Castaño et al. (2001b) who expressed that some environments can be more or less favorable in the expression of white rot symptoms than others.

Differences among the 47 sunflower genotypes and inconsistency of relative hybrid performance were detected in Camet and Balcarce after *S. sclerotiorum* infections on capitula. Non-controlled variability values agree with the level of precision reported by Vear and Tourvieille (1988) and Álvarez et al. (1999) when a set of sunflower commercial hybrids was evaluated on white rot incidence using a similar protocol of infection to that of E3. The environmental effects affected the magnitude and expression of DI and SAI traits in sunflower hybrids. Therefore, these results indicate that experiments should be conducted in different environments in order to detect genotypes with good performances to DI and SAI. However, the ranking of genotypes did not significantly change from one location to another as the differential responses of hybrids compared to others were a matter of scale. This type of

GE interaction is known as non-crossed or quantitative interaction (Baker, 1988). Non-crossed GE interactions were also reported by Tourvieille et al. (1996), Hahn (2002) and Serre et al. (2004) when sunflower responses to white rot were evaluated under European conditions, which are quite different to those ones that are normally present in Camet and Balcarce. In addition, the genetic materials involved in the trials of Europe and Argentina are not genetically related to each other. So, even when those environment and genetic conditions were distinct in France, Germany and Argentina, the results obtained in these studies were similar in relation to the effects and type of GE interaction, and therefore it suggests that this type of GE interaction could be characteristic of white rot disease.

When selecting genotypes for a wide adaptation area, as it could be the region of the province of Buenos Aires in Argentina, sunflower breeders prefer either no GE interaction or non-crossed nature, otherwise the selection for a broad adaptation would be more difficult and would result in reduced response to selection (de la Vega, 2004). Fortunately, the results of our work indicated that non-crossed GE interaction is more important. Then, testing programs and selection for white rot resistance could be facilitated as it would not be necessary to define neither the narrow range of environments nor the sowing conditions for adapted sunflower hybrids.

Although several papers have described the sunflower resistance to either DI or SAI (Vear and Tourvieille, 1987, 1988; Castaño et al., 1989, 1993; Álvarez et al., 1999), none of them has identified genotypes showing both favorable attributes. In this sense, the high variability of hybrid responses at each resistance factor detected in the present work, and always higher than the one calculated for the GE interaction, would greatly help in selecting the most superior hybrids and discarding the susceptible ones to white rot resistance.

The presence of non-crossed GE interactions allowed the selection of four hybrids (DK3×R199, DK6×R199, SD×R199, and SD×PAC1) with low DI values at Camet (E1) and Balcarce (E3); all these hybrids were placed in the G3 group at each location. When the level of resistance of these hybrids was compared to that one shown by the same genetic materials but evaluated under natural infection (E2) only two (DK3×R199 and DK6×R199) had the same high level of resistance, therefore validating their high performance. The high level of data precision reached under artificial infection as well as the positive correla-

tion between artificial and natural infection determined that selection by hybrid performance was made in that order.

The second resistance variable analyzed in our study was SAI. The selected hybrids DK3×R199 and DK6×R199 showed a similar incubation period in relation to the hybrid SD×PAC1, well-known in reducing the pathogen mycelium growth in the capitula tissues according to Vear and Tourvieille (1987, 1988) and Castaño et al. (1993), and higher values than the moderately SCKR resistant check hybrid. In this sense, the appearance of first symptoms would be therefore retarded in those selected hybrids.

Most of the evaluated hybrids in this work were obtained by crossing elite inbred lines used to develop sunflower hybrids with a high yield potential of seed and oil. Therefore, the sowing of the hybrids DK3×R199 and DK6×R199 would allow them to have a good agronomic performance under favorable conditions to *S. sclerotiorum* attacks on capitula in the sunflower grown area of the province of Buenos Aires in Argentina.

Although this work showed that two hybrids performed well in three environments of the most important sunflower growing region in our country, a stability study using as variables other environmental conditions (under dry situations, for example) could help to determine whether these genetic materials show similar level of resistance to white rot and then could be sown in the wide Argentinean sunflower growing area without increasing the disease risk.

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